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**Cover:** Tiger rattlesnakes, *Crotalus tigris*, in courtship, 4 September 2010, Pinal County, Arizona. Photograph by Hans-Werner Herrmann.


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# Notes on the Herpetofauna of Western Mexico 24: Predation on *Lithobates neovolcanicus* (Hillis & Frost, 1985) by *Quiscalus mexicanus* (Gmelin, 1788) in the Gardens of Guadalajara Zoo, Jalisco, Mexico

**Daniel Cruz-Sáenz [1], David Lazcano [2], José Orlando Ríos-Martínez [2], Juan Antonio García-Salas [3], Andrés Rodríguez-López [1] and Larry David Wilson [5]**

## Abstract

In this note we report an incident in which two sympatric species were involved, one the great-tailed grackle as predator (*Quiscalus mexicanus*), and the other the neovolcanic leopard frog (*Lithobates neovolcanicus*) as prey. We also document some aspects of the biology of each species as components of their trophic chain.

## Resumen

En este documento reportamos un incidente donde participaron dos especies simpátricas uno como el depredador zanate Mexicano (*Quiscalus mexicanus*), y el otro como la presa rana neovolcanica leopardo (*Lithobates neovolcanicus*), además documentamos algunos aspectos de la biología de cada especies como componentes de la cadena trófica.

## Introduction

We document predation on *Lithobates neovolcanicus* (neovolcanic leopard frog) by *Quiscalus mexicanus* (great-tailed grackle) and discuss some aspects of the biology of these two inhabitants of the grounds of the Guadalajara Zoo in Guadalajara, Jalisco, Mexico.

## Background on *Lithobates neovolcanicus* (Hillis & Frost, 1985)

The neovolcanic leopard frog has been registered in the states of Aguascalientes (Vázquez-Díaz and Quintero-Díaz, 2005), Guanajuato, Michoacán (Hillis and Frost, 1985; Flores-Villela and Canseco-Márquez, 2007), Jalisco (Hillis and Frost, 1985, García and Ceballos, 1994; Flores-Villela and Canseco-Márquez, 2007, Reyna-Bustos et al., 2007; Godoy González, 2014), Zacatecas (Ahumada-Carrillo, 2010) and México (Aguilar-Miguel et al., 2009).

The neovolcanic leopard frog reaches a snout–vent length of approximately 10 cm. The head is triangular in appearance. The eyes are large in size, and on the body are two dorsolateral folds in the form of yellow stripes that run from the nuchal area to the groin. This frog's dorsal ground coloration varies from green to brown with dark spots scattered irregularly about. The belly is yellow-white (Vázquez-Díaz and Quintero-Díaz, 2005; Cruz-Sáenz et al., 2008).

*Lithobates neovolcanicus* adults are semiaquatic, living on the shores of both intermittent and permanent bodies of water. They are mainly crepuscular and nocturnal, but sometimes can be found active during the day. Breeding occurs during spring and summer (Vázquez-Díaz and Quintero-Díaz, 2005). Reproduction is oviparous with indirect development. Males vocalize during the breeding season on the banks of water bodies, mainly during the night. The eggs are deposited in aquatic vegetation forming a spherical mass, which can contain about 4500 embryos; each egg is 1.7 mm in diameter with a 4-mm-diameter gelatinous covering (Hillis and Frost, 1985; Reyna-Bustos et al., 2007). Also important to mention is the finding of malformed individuals (Barragán-Ramírez and Navarrete-Heredia, 2011). The adults feed on small insects they find on the edges of water bodies (Vázquez-Díaz and Quintero-Díaz, 2005). A specific diet item was documented by González Hernández et al. (2010), who found a *Lithobates neovolcanicus* eating an *Anaxyrus punctatus* (red-spotted toad) in a small pond situated at Río Las Lajitas in the Tortugas locality at Xichú Municipality, Guanajuato, Mexico. Tadpoles are seen during most of the year, suggesting a long breeding season. Dixon and Lemos-Espinal (2010) mentioned that the larvae can remain in the water for a long period of time (a year or more), depending on the environmental conditions. There is no detailed information on the species' diet, except what was documented recently by Cruz-Sáenz et al. (2019), who found a group of *L. neovolcanicus* tadpoles feeding on a mango fruit (*Mangifera indica*) that had fallen into an artificial pond.

Concerning its conservation status, this frog is categorized as follows: on the Mexican Wildlife list (SEMARNAT, 2010) as Threatened (A); on the IUCN Red List (IUCN, 2020) as Near Threatened (NT); with an Environmental Vulnerability Score in the Medium range (13) (Cruz-Sáenz et al., 2017). Unfortunately, this species has been detected as positive for *Batrachochytrium dendrobatidis* (Frías-Alvarez et al., 2008; Mendoza-Almeralla et al., 2015; Nava-González et al., 2020), which could increase the decline in their populations.

1. Universidad de Guadalajara, Centro de Estudios en Zoología, Centro Universitario de Ciencias Biológicas y Agropecuarias, Apartado Postal 1-1919, Guadalajara, Jalisco, C.P. 44101 México. dcruzsaenz@gmail.com. (DCS); andres.rlopez@alumnos.udg.mx (ARL)

2. Universidad Autónoma de Nuevo León, Facultad de Ciencias Biológicas, Laboratorio de Herpetología, Apartado Postal 157, San Nicolás de los Garza, Nuevo León, C.P. 66450 México. imantodes52@hotmail.com. (DL)

3. Universidad Autónoma de Nuevo León, Facultad de Ciencias Biológicas, Laboratorio de Ornitología, Ciudad Universitaria s/n, San Nicolás de los Garza, Nuevo León, C.P. 66450 México. juan_0305@hotmail.com.

4. Centro Zamorano de Biodiversidad, Escuela Agrícola Panamericana Zamorano, Departamento de Francisco Morazán, Honduras; 1350 Pelican Court, Homestead, Florida 33035-1031, USA. bufodoc@aol.com.

## Background on *Quiscalus mexicanus* (Gmelin, 1788)

The great-tailed grackle, native to the Gulf of Mexico slope, is currently distributed throughout the country (Christensen, 2000, Gurrola-Hidalgo et al., 2009), as there are records in Hawaii, Canada, the United States of America, Central America and the Caribbean, as well as in Venezuela, Colombia, Ecuador, and Peru in South America (AOU, 1998). The broad distribution of this bird is due to its adaptability and tolerance to environments modified by humans (Wethje, 2003; Gurrola-Hidalgo et al., 2009). This species has a broad diet, including many vertebrates, invertebrates, small crustaceans and other marine animals, as well as grains and fruits (Gurrola-Hidalgo et al., 2009). Prey items may include Orthoptera (crickets and grasshoppers), Coleoptera (beetles), Lepidoptera (butterflies and moths), Hymenoptera (bees, wasps, and ants), Diptera (flies), Homoptera (aphids and cicadas), arachnids (spiders), gastropods (snails and slugs), as well as different types of crabs, such as Beebe's fiddler crab (*Uca beebei*) and shrimp (Scutch, 1954; Davis and Arnold, 1972; Oberholser, 1974; Johnson et al., 1989; Peer and Bollinger, 1997; Grabrucker and Grabrucker, 2010; Johnson and Peer, 2020). These birds also consume a wide variety of vegetable materials, such as corn (*Zea mays*), rice (*Oryza sativa*), sorghum (*Sorghum bicolor*), oats (*Avena saliva*), grapes (*Vitis* sp.), figs (*Ficus* sp.), berries, tigernut tubers, food in garbage dumps, carrion, and even bananas.

The specific documentation of its vertebrate diet includes various anuran tadpoles, brown anole (*Norops sagrei*), Sonoran spotted whiptail (*Aspidoscelis sonorae*), Mediterranean house gecko (*Hemidactylus frenatus*); green iguana (*Iguana iguana*), southern black-nosed lizard (*Sceloporus melanorhinus*), western fence lizard (*Sceloporus occidentalis*), neonates of olive ridley sea turtle (*Lepidochelys olivacea*), barn swallow (*Hirundo rustica*), and chicks of the white-winged dove (*Zenaida asiatica*); it is also known as a thief of the nestlings of other birds (Gurrola-Hidalgo et al., 2009; Sánchez-Soto, 2015; Cupul-Magaña et al., 2018).

## Background of the study site

The Guadalajara Zoo is a decentralized public body administered by the Guadalajara city council. Located in the "Barranca de Huentitán," it emerged from the need for Jaliscan society to have a space for leisure and recreation, as well as a space that would inspire interest for the conservation of nature. The zoo was inaugurated on March 11, 1988. On the 25th of that same month, it opened its doors to the general public. This facility is one of the few self-financing zoos in Mexico. The zoo has 50 fully-developed hectares for various wildlife exhibits. The topography of the area supports natural and ornamental flora. Also within the grounds are 280 hectares of ecological reserve.

The Guadalajara Zoo adjoins a natural protected area of the state called "Barrancas de los Rios Santiago y Verde" that is in the physiographic region of the Sierra Madre Occidental, according to Cruz-Sáenz et al. (2017). The faunal diversity of the area still has been poorly documented, but local researchers have undertaken inventory studies of the flora and fauna of this particular canyon, resulting in lists and field guides of the local fauna, such as the report of 58 species of arthropods (Navarrete-Heredia et al., 2008), nine amphibians and 35 reptiles (Cruz-Sáenz et al., 2008), 70 birds (Maya-Elizarrarás et al., 2008), and 47 mammals (Godínez-Navarro et al., 2008). Other papers dealing with the herpetofauna of the Río Santiago Canyon include one on the herpetofauna of Huaxtla (Cruz-Sáenz et al., 2011), one on the sympatric species of *Xantusia sanchezi* at Huaxtla (Cruz-Sáenz et al., 2010), one on the herpetofauna of the municipality of Hostotipaquillo (Flores-Cobarrubias et al., 2012), and a book on the wildlife near Guadalajara (Ponce-Campos and Huerta-Ortega, 2004).

## Methods and Results

Here we document a new food item for the great tailed-grackle (*Quiscalus mexicanus*) in a photographic sequence of the event (Figure 1). We observed the incident while visiting the zoo in

**Figure 1**. 1–4) Photographic sequence of a male great-tailed grackle (*Quiscalus mexicanus*) feeding on a neovolcanic leopard frog (*Lithobates neovolcanicus*) in the garden grounds of Zoológico de Guadalajara, Guadalajara, Jalisco, Mexico. Photographs by Daniel Cruz-Sáenz.

**Figure 2**. Dorsal and ventral views of the *Lithobates neovolcanicus* prey item. Photographs by Daniel Cruz-Sáenz.

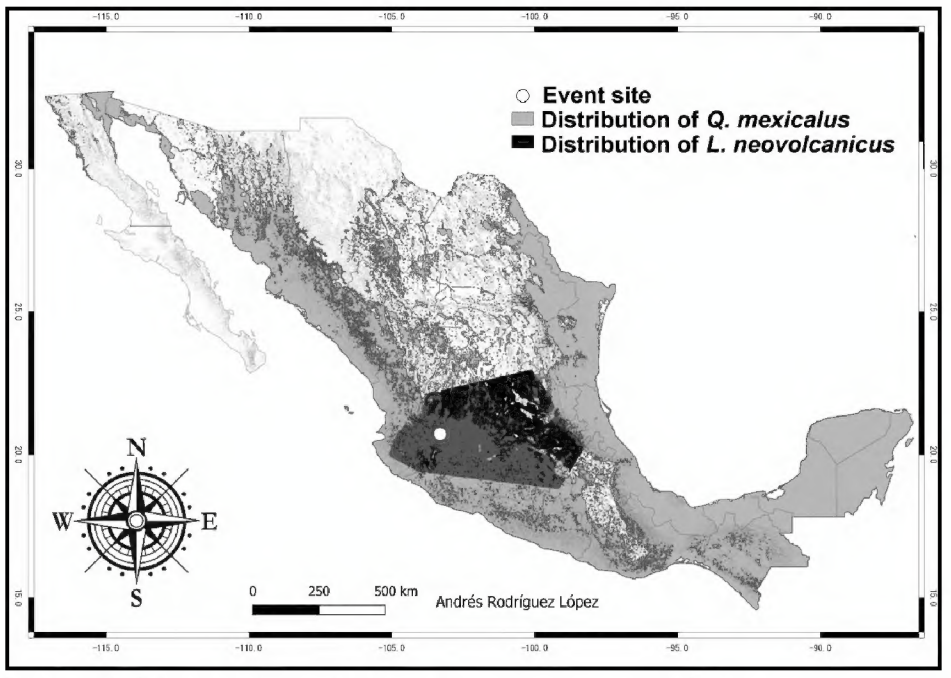


**Figure 3**. Distributions of *Quiscalus mexicanus* and *Lithobates neovolcanicus* in Mexico.

connection with a lecture on the exhibition of some of the most popular vertebrates there, with a group of students from our Faculty of Biological Sciences of the Universidad de Guadalajara. On Wednesday 15 May 2019, in the Guadalajara Zoo gardens near the aquarium adjacent to a large pond in which some spider monkeys are displayed on an island, we found a great tailed-grackle *(Quiscalus mexicanus*) male eating an adult individual of the neovolcanic leopard frog (*Lithobates neovolcanicus*). The event occurred at 2:54 P.M. The grackle was positioned on a broken branch at ground level; when we moved closer, we discovered a neovolcanic leopard frog being eaten by this bird. Initially, it was not bothered by our presence. The event lasted for approximately 33 minutes. We believe that this frog came from the nearby pond, as we observed other individuals in that area. We do not know if the frog was captured in the garden or in the pond. The vegetation of the area is composed of native trees, eucalyptus trees, and a few bougainvilleas.

## Discussion and Conclusion

Of the 10,500 bird species in the world, Mexico has between 1123 and 1150 wild species, representing approximately 11% of the world total. Of these, 77% are permanent resident species; the rest are winter-visiting species and 87 species that are migratory, rare or accidental. Between 194 and 212 species are endemic to Mexico, representing 18–20% of those registered for the country. Between 298 and 388 species (26–33%) are in some category of risk. There are 10 species introduced by man and 4 species extinct in the twentieth century (Scutch, 1954; Navarro-Sigüenza et al., 2014). In particular, the family Icteridae is represented by 98 species in the world, with 35 present in Mexico. Besides the great-tailed grackle, this family includes the common grackle (*Q. quiscala*), which has been reported to include frogs in its diet (Davis and Arnold, 1972). All icterids are omnivorous. Over much of its range *Lithobates neovolcanicus* is sympatric with *Quiscalus mexicanus* (Figure 3). We can assume that predation events such as the one described herein happen regularly where the species are sympatric. These types of events are poorly documented. We encourage herpetologists and other naturalists to keep their eyes open whenever they undertake fieldwork.

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# Anthropogenic Drivers and Chytridiomycosis: Untangling the Disappearances of the Golden Toad and Costa Rican Variable Harlequin Toad and Addressing Amphibian Decline

**Jeffrey L. Coleman**
**732 S. Financial Place,**
**Chicago, IL 60605**
**jeffcoleman844@berkeley.edu**

## Introduction

In the last few decades, the world has faced an unprecedented decline in its amphibian populations. By 2005, at least 43% of approximately 7,000 discovered amphibian species had experienced population declines, 32.5% were globally threatened, 427 were critically endangered, 34 were extinct, and 122 were claimed by the Global Amphibian Assessment (GAA) to possibly be extinct (Pounds et al., 2006). Among the vertebrates, the class Amphibia has the most species represented in the endangered and critically endangered categories. Some of the most rapidly declining species are found in upland Neotropical and Paleotropical riparian habitats, frequently in areas that are protected. Disappearances here are often said to be enigmatic due to the lack of an obvious cause (Lips et al., 2006). Among these mysterious losses are the extinctions in the late 1980s of the golden toad (*Incilius periglenes*), formerly abundant in the Monteverde Cloud Forest Preserve and endemic to Costa Rica, and the Costa Rican variable harlequin toad (*Atelopus varius*), which became extinct locally in the Monteverde region around 1989, and has since declined drastically across the country. Of these two species of true toads (family Bufonidae), only one remaining population of the latter is known today (Pounds and Crump, 1994; González-Maya et al., 2013). In this review, I explore current thinking as to the principal causes of these losses, and what research-driven and policy-based solutions scientists have devised to address both the population decline of tropical amphibian species like *A. varius* as well as the global amphibian decline (GAD).

## Main Causes of Golden Toad and Harlequin Frog Losses

It is exceedingly likely that *Incilius periglenes* has gone extinct, as it has not been rediscovered in the wild since 1989. Two theories proposed in 1994 to explain the extinction of *I. periglenes* were either that this species could be experiencing

natural fluctuations in its population, or that this species might be present in underground retreats in anticipation of better breeding conditions. Results from the Pounds et al. (1997) evaluation of probabilistic null models incorporating the number and severity of disappearances of amphibians in Monteverde, including that of *I. periglenes*, suggested losses significantly beyond those reflecting standard demographic variability. Forty percent of anuran fauna had declined throughout the authors' 1990–1994 surveys, whereas other organisms had not declined to a similar extent, implicating anthropogenic causes. In addition, sightings of *I. periglenes* and *A. varius* dropped by about 99% in the same year. This indicates high adult mortality rather than just unsuccessful breeding and recruitment; adult survivorship in the bufonids had always been higher than 1% during declines (Pounds and Crump, 1994). These issues make the population flux hypothesis unlikely. The underground retreat hypothesis is also an unlikely explanation for the loss of *I. periglenes*. *Favorable* weather conditions have occurred since 1987; these would likely have drawn the amphibians out from hiding (Pounds et al., 1997).

The extinction of *I. periglenes* has also been attributed to extreme weather conditions, ultraviolet radiation, microparasites, atmospheric contamination, and deforestation. There could have been an acid rain event in 1987. However, tests in subsequent years showed no atypical acidity levels, and while high acidity has been shown to affect the distribution of terrestrial adults and to kill aquatic embryos, no link has been shown with high adult mortality. It is possible that ultraviolet radiation could kill adults of *A. varius*; however, *I. periglenes* hide in retreats about 95% of the time, and upon emergence, are often protected by cloud cover under a dense forest canopy. Deforestation and habitat loss appear unlikely as key drivers as well; it has been shown in studies that, when controlling for these factors, they cannot explain the widespread extinctions and declines of either *A. varius* or *I. periglenes* in computational estimates (Pounds et al., 2006). In studies of parasites in 1984, it was shown that a variety attack amphibians, including viruses, bacteria, and protozoans. However, it is improbable that these suddenly became fatal to *A. varius* and *I. periglenes* without interplay with climate or another driver (Pounds and Crump, 1994).

Pounds and Crump (1994) also investigated the abnormally warm, dry conditions of 1987: in addition to temperature reaching a record high, there was abnormally low rainfall in the periods referred to as the late wet season, the transition into dry season, the dry season, and the post-dry season. Also, there was record low flow in local aquifer-fed streams during the dry and post-dry seasons. These anomalous climate change events were associated with the 1986–1987 El Niño/Southern Oscillation; the effects of this event were more severe than those associated with the more powerful El Niño of the years 1982 and 1983. These findings led the authors to describe two hypotheses: the climate-linked epidemic hypothesis and the climate-linked contaminant pulse hypothesis.

The investigators found that suboptimal conditions affecting vitality and energy budgets could lead to high adult mortality by changing the outcome of critical biological interactions. In the case of *A. varius*, warm, dry conditions can lead to habitat patchiness and thus to a highly clumped dispersion pattern. With respect to parasites, this decrease in inter-host distance and increased density can facilitate an epidemic. Given this hypothesis, it was viable that either a "non-species specific pathogen" could have caused the decline of amphibians at Monteverde or that different microparasite species could have impacted the various amphibian populations. These findings are the basis for the climate-linked epidemic hypothesis.

The climate-linked contaminant pulse hypothesis is supported by evidence of long-range contamination that occurs in Monteverde; a visible haze in that occurs in March–April results from increased atmospheric turbidity. This can be explained by temperature inversions at 2000–3000 meters, wind erosion of soils, and agricultural burning in the lowlands. In addition, cloud water collected during these months contains especially high levels of phosphates and nitrates. Atmospheric contamination caused by excessive use of insecticides, herbicides and fungicides also occurs here, due to volatilization, wind erosion of deposited residues, and drift during chemical application (Pounds and Crump, 1994). A climate disturbance could interact with this contamination in a multitude of ways; for example, during a dry spell, less precipitation would lead to a higher concentration of pesticides and chemicals in the soil. Also, the body of a rehydrating amphibian may re-absorb only 60–70% of the amount of water equivalent to its normal body water content, meaning it would have less body fluid to dilute toxic compounds (Putnam and Hillman, 1977). The patchy nature of amphibian declines in mountainous regions could reflect the location of contaminant sources and the effect of topography on cloud-flow patterns and wind-currents (Pounds and Crump, 1994).

While contamination is potentially a factor in driving environmental stress and leading to amphibian disease outbreaks, the fungal disease known as chytridiomycosis, as well as changing climate, is currently considered the most probable cause of the declines of *A. varius* and *I. periglenes* (Shunthirasingham et al., 2011). Chytridiomycosis is an emerging infectious disease caused by a possibly saprophytic pathogen called *Batrachochytrium dendrobatidis* (*Bd*). Field as well as laboratory work suggest that the disease, which grows on the skin, spreads by amphibian-to-amphibian and environment-to-amphibian contact via zoospores. Animals at cooler temperatures shed zoospores more slowly, and zoospores appear at lower density in water. Amphibians can carry the infection latently, and can live up to 220 days even if the disease is symptomatic. Lips et al. (2006) suggested that chytridiomycosis is the leading cause of global amphibian extinction; chytridiomycosis proved to be an epizootic with high disease-induced mortality when studied on an uninvaded site. The timing of the outbreak suggested its movement was southeastward in Central America, first in Costa Rica and then into Panama.

Specimen-based research conducted in the Museum of Vertebrate Zoology at the University of California, Berkeley, corroborated other findings of a temporal and spatial spread of this disease. *Bd* was absent in all countries prior to its first detection, contradicting the prevalent school of thought that the pathogen was present in the environment and emerged in response to climate change. This had never been shown before; this study

determined that the sole cause of *Bd* spread is host densities surpassing a critical threshold. However, this research did not rule out the role of climate change as an agent in Monteverde or throughout Central America, though climate change did not appear to play a role in the extinction events at the sites studied (Cheng et al., 2011). Further, the case could be made that, even within the context of this study, increased host densities and resulting pathogen virulence could be linked to climate change. Pounds and Crump (1994) posited that the warm and dry conditions of 1987 forced members of *A. varius* of the Rio Lagarto population in Costa Rica to abandon their crevices and congregate in wetter areas closer to the stream. It has also been shown that *Bd* thrives under moist, cool, conditions, an observation that explains the sensitivity high-elevation rainforest amphibians have had to pathogen-induced declines (García-Rodríguez et al., 2011). Thus, increased host density in wet retreat sites during times of drought and warmth increases amphibian susceptibility to the recently arrived *Bd*; then, proliferation and virulence are potentially favored during subsequent cool temperature cycles (Longo et al., 2010).

Pounds et al. (2006) offered alternative hypotheses to explain how *Bd* became so lethal during the warm, dry season in light of the experiments that demonstrated *Bd*'s success in moist, cool conditions. Stress from warm and dry years may increase amphibian vulnerability to chytridiomycosis, or warm climates could favor the pathogen directly.

Both low and high altitudes seem to limit the effect of *Bd*. Pounds et al. (1999) used daily rain-gauge records to develop a mist-frequency index. Reduced mist frequency is associated with shifts in vertebrate populations and with the disappearance of *A. varius* and *I. periglenes* in the Monteverde cloud forest. Regional climate trends were examined to discern how sea surface temperatures (SST) and air temperatures (AT) related to local tendencies on differing scales. Extinctions have been associated significantly with AT and SST, and thus with warming, for the tropics (Pounds et al., 2006).

As a general rule, global warming increases the rate of evaporation as well as the air's capacity to sustain water; as water vapor rises and condenses, latent heat is transferred to the atmosphere. With the help of condensation nuclei from aerosols, increased water vapor can lead to increased cloud cover. Cloud cover increases nocturnal warming by reducing heat loss at night. Cloud cover can also reverse daily trends in climate by obstructing solar radiation, which often diminishes warming throughout the day. This is evident in Monteverde, where the daily minimum temperature is rising and the daily maximum is falling during all four seasons (Pounds et al., 2006). Daily temperature range decreases for both wet and dry days with increased cloud cover (Pounds et al, 1999).

Increased cloud cover should favor the chytrids; increased cloud cover creates moist conditions and shields the fungi from excessive warmth. Microscale cooling can overshadow local ambient trends, which should also promote chytrid growth. If amphibians want to find warmth to fight infection, heightened cloudiness might thwart their defenses.

The chytrid-thermal-optimum hypothesis, put forth in light of the pattern of *A. varius* declines, posits that the lowlands are frequently too warm for the fungus during the day and the highlands too cool for it at night. Vertical thermal profiles have suggested a moist adiabatic lapse rate whereby the drop in temperature with increasing elevation had lessened in the years leading to this species' most intense declines. This daytime cooling in tandem with nighttime warming increases disease development rate, and most extinctions have had occurred in places where the minimum temperature is shifting toward the growth optimum for *Bd*. Greenhouse warming and the intensification of the hydrological cycle, along with aerosol pollution, influence the patterns of cloud formation and change the thermal, photochemical, and moisture environments of many organisms. This alters ecosystem balance and hurts species' chance for survival (Pounds et al., 2006).

Pounds and Masters (2009) defended the validity of the chytrid-thermal-optimum hypothesis. This paper is implicitly opposed to the findings of the Museum of Vertebrate Zoology study (Cheng et al., 2011), which did not find climate change to be a necessary component in the lethality of *Bd*. Evidence suggesting that climatic changes occurred before chytridiomycosis outbreaks or even before *Bd* arrival does not cast doubt on the chytrid-thermal optimum hypothesis; the notion that climate change favors the temperatures that support this newly arrived fungus, is, on the contrary, reconcilable with the hypothesis.

**Targeting Amphibian Declines through Research**

Work in comparative physiology has helped us understand the relationship between chytridiomycosis and temperature change. It has been clear for years that the relationship between these infectious agents and their frog and toad hosts is mediated by thermoregulatory behaviors and thermal ecology. Temperatures well above what support the fungi that are only moderate for toads and frogs can allow these amphibians to fight infection. We know now that anurans can thermoregulate and help their bodies survive bacterial infection. Even so, to understand why anurans are still vulnerable to chytridiomycosis, it is imperative to examine field body temperatures, microhabitat selection, and patterns of activity. In addition, we need to delve into physiological ecology to understand, for example, how toxic environmental conditions may be lowering the protection derived from anuran skin products and increasing the levels of circulating glucocorticoids, which may be damaging in high concentrations. The skin secretions and immune response of an anuran also depend on their local environment; alkaloids are sequestered from rich food sources such as millipedes and ants. The quality of the microenvironment can help or harm an amphibian's biochemical profile (Navas and Otani, 2007).

There are several broad research objectives aimed at curtailing GAD, which focus on the chytridiomycosis side of the fungus-climate synergy. Research in this area has distinguished between lowland species of frogs, particularly of the genus *Atelopus*, and upland species, as we suspect *Bd* interacts with these species and the environment variably along elevational gradients (La Marca et al., 2005). In addition, Longo et al. (2010) urge us to examine the effect of *Bd* on persistent populations, *Bd* movement patterns and interactions, defense mecha-

nisms, and disease physiology.

We can boast of several visible results that portend success. By 2004, advances in disease physiology meant that detection of *Bd* was now superior, with a rapid, non-invasive, sensitive PCR-based assay called the TaqMan assay. This assay, which had proven to be more reliable than the previous convention of performing histology with toe clippings, is a powerful tool for prohibiting the spread of *Bd*. It has allowed for the implementation of quarantine regimes, and early disease detection can help elicit conservation efforts for endangered species, as well as the repopulation of captive breeding populations into depopulated habitats. On a larger scale, this assay could contribute to a better understanding of the pathogen and to stopping its spread into new territories (Boyle et al., 2004).

*Atelopus elegans* could be a model species to better understand how *Bd* could be effectively inhibited, as it has skin bacteria that provide high anti-*Bd* action. Nonetheless, further research is necessary to determine how bacterial symbionts and consortia interact with each other and the amphibian skin. Probiotic therapy has been suggested as a management tool to target the protection of Neotropical amphibian species from the effects of *Bd*; applying microbiota as treatment has proven effective as a defense in temperate zone amphibian species (Flechas et al., 2012). Antimicrobial peptides, housed in dermal granular glands and secreted into the mucus as one of the first lines of defense following alarm, have shown more promise in non-declining rainforest amphibian species than in declining ones. This defense method against skin pathogens like *Bd* relies on a wide array of genes that express these peptides. However, these peptides must also be able to harmonize with potentially beneficial microbiota, such as those on the *A. elegans*. More genetics research needs to be conducted to better understand their function (Rollins-Smith, 2009).

Goals on the climate side of the synergy emphasize aggressive research on amphibian thermal tolerances and preferences, dehydration breadths, and the opportunity costs of water conserving behavior. In addition, we still know little in regards to the actual moisture and temperature ranges experienced by amphibians. One factor limiting climate change research is that it is nearly impossible to model in the field, and the effects of climate change will not be fully known until it intensifies further.

In understanding the climate-fungus synergy as a whole, a review by Li et al. (2013) cautioned that additional rigor is needed in shifting to more deductive and parameter-estimation type models that are designed to narrow down the plethora of hypotheses. This would entail "correlative field studies, experiments on extant 'model' amphibian species, and mathematical models and interdisciplinary collaborations to build a weight-of-evidence case for a causal association between climate change and amphibian declines." As a whole, this paper pushed for functional, physiological-based models in conjunction with field data on climate. Collecting the requisite data will take time.

Researchers have also suggested conservation methods aimed at treating the population loss symptoms of Neotropical amphibians directly, not just those tackling the environmental phenomena underlying these amphibian declines. A recent survey in Costa Rica examined amphibian population relicts, species that inhabit a vastly smaller geographic range than they did previously, usually due to environmental or climate change. Using a maximum entropy algorithm, it was found that regions with high likelihood for relictual populations corresponded with areas with recently rediscovered species: for example, in Monteverde, *Duellmanohyla uranochroa* and *Isthmohyla rivularis* were rediscovered, in addition to populations of *Lithobates vibicarius*. The potential discovery of relictual populations in secondary or fragmented forests will hopefully incentivize improvements in management or restoration of these habitats. These regions could be used as hot spots for more research on the climate-fungus synergy by combining the methods of this survey with physiological niche modeling. These computational methods, which greatly increase the chance of finding and being able to rigorously monitor relictual populations could, moreover, be used to study these communities themselves and elucidate why these populations persisted while other conspecific ones have expired. This will also help us determine how to conserve them for generations to come. On a grander scale, these types of methodologies could be used to conduct a census of amphibian populations worldwide and rediscover lost amphibians (García-Rodríguez et al., 2011).

Several captive breeding programs have been established, including for a population of *A. varius* found in Panama. Successful programs at the Baltimore and Detroit Zoos have kept and bred populations of *A. varius* and *A. zeteki*. Currently, captive breeding is considered to be the strongest method to maintain wild populations that do not combat root causes, such as climate change and *Bd* spread. The authors of La Marca et al. (2005) are resigned to the idea that, in wild populations with factors that are difficult to control such as climate change and *Bd*, the most effective conservation tool is *ex situ* breeding.

A 2014 study analyzed this type of *ex situ* conservation with *Atelopus* frogs, studying group housing strategies. This analysis, the first ever to use fecal glucocorticoid levels to measure stress response in these frogs, found that males interacted with each other combatively at first, which was correlated with higher stress. However, after a few weeks, the frequency of these negative interactions lowered, representing a drop in stress hormones. This paper has powerful ramifications for efforts in amphibian conservation and reintroduction. Further studies need to be conducted to determine optimal housing densities and to discover if group housing, in the long-term, leads to increased gut parasite load, changes in body conditions when more submissive males do not compete as well for food, or relapses in aggressive interactions during the breeding season. These findings would then call for intense monitoring and careful consideration before changes in management are made (Cikanek et al., 2014).

**Targeting GAD through Policy**

The first meeting of the World Congress of Herpetology in England in 1989 and a meeting in the United States the following year led to the creation of the Declining Amphibian Populations Task Force (DAPTF), under the patronage the IUCN Species Survival Commission (SSC), in 1991. This establishment served to research the causes and severity of the declines.

Small herpetology groups were dispatched worldwide as a result, and this stimulated the creation of the *Froglog* newsletter to keep enthusiasts informed about advancements in conservation and research. By 2004, in the aftermath of these papers and projects, the IUCN was prompted to conduct the GAA, whose job it was to focus on data deficient species in clarifying biodiversity, amphibian decline, and ecological information.

An Amphibian Conservation Summit ensued in Washington, D.C., in 2005, followed by the publication of the IUCN SSC's Amphibian Conservation Action Plan (ACAP). This plan identified key issues required to curb the amphibian crisis, and provided an elaborate framework around intervening, which included an estimated US budget of around 400 million dollars applied between the years 2006 and 2010. This would be sponsored by an Amphibian Action Fund (AAF) that stakeholders in the issues planned to initiate (Bishop et al., 2012). This action plan, which described itself as "the most ambitious program ever developed to combat the extinction of species," detailed four suggestions for curbing amphibian decline: "1) expanding our understanding of the causes of declines and extinctions, 2) continuing to document amphibian diversity and how it is changing, 3) developing and implementing long-term conservation programmes, and 4) responding to emergencies and immediate crises" (Gascon et al., 2007).

In addition to advocating for fundamental research, the first of the four suggestions called for the establishment of Regional Centers for Disease Diagnostics around the world. Their job would be to manage and direct free disease testing for field research groups trying to curb amphibian decline. Funding of these clinics would come from the AAF. In addition, rapid response teams would be started to treat disease outbreak early in its occurrence.

The plan recommended that the GAA have a team devoted to updating it and making species data available continuously to better keep track of amphibian diversity. In addition, the GAA was to maintain its website, make data reporting altogether more reliable in challenging areas, more rigorously distinguish between real species decline and ostensible declines resulting from inexhaustive sampling, and share its discoveries.

For objective three, the ACAP planned to implement a program specifying the 120 top priority conservation regions and conduct management plans, monitoring, assessment, and funding searches at each. Another program in reintroduction would be established once disease management was sufficiently sophisticated, in which people trained in reintroduction would work for both the captive bred and wild amphibian program. The last program for this objective would be in harvest management. This would aim to create sustainable harvest programs, promote wildlife law enforcement, oversee amphibian harvest and trade, institute recovery programs for impacted species, ensure captive breeding programs are certified, and improve public awareness around the role of harvest and trade in many amphibian species' decline around the world.

The last objective called for the development of an emergency response plan, activated through regional response teams. These teams would be trained to immediately address events such as an emergent disease outbreak. The techniques behind addressing such an emergency would be developing emergency captive breeding colonies within the countries of origin, developing short and long term captive management plans, training and capacity building within the range country, supporting and developing captive breeding sciences, supporting research into disease management, and supporting educational and outreach programs. The Amphibian Emergency Fund would sponsor the conservation of remaining habitats in the top priority sites. These immediate conservation measures would conserve remaining habitat fragments that provide for communities of species at risk of extinction. Species on the brink of extinction due to over-harvesting would also be addressed through these emergency funds (Fenolio, 2009).

In June of 2011, the Amphibian Survival Alliance (ASA), was founded. As a global partner in amphibian conservation with clout over a consortium of conservation organizations worldwide, it is in a strong position to implement the ACAP. Its objective is to restore all threatened native amphibian species to their healthy population levels and into their natural roles worldwide. The ASA also has new initiatives for tackling the ACAP issues, which include the creation of a web-based "living" version of the ACAP document, as well as an attempt at what they claim will be a more collaborative approach in implementing the ACAP themes (Bishop et al., 2012). These actions, if put into place, would help curb one the most pressing threats to the biological and ecological diversity on planet Earth: the disappearance of amphibians.

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# Notes on Reproduction of Plains Leopard Frogs, *Lithobates blairi* (Anura: Ranidae), from Oklahoma

**Stephen R. Goldberg**
**Whittier College, Biology Department**
**Whittier, CA 90608**
**sgoldberg@whittier.edu**

## Abstract

I conducted a histological examination on gonadal material from 49 adult and one subadult *Lithobates blairi* from Oklahoma. The smallest mature male (sperm in lumina of seminiferous tubules) measured 53 mm SVL and was from November. The smallest mature female (mature oocytes) measured 57 mm SVL and was from September. Varying amounts of follicular atresia (spontaneous degeneration of oocytes) was noted in seven (22%) of the 32 adult females in the sample. The ovary of one female from June contained postovulatory follicles (evidence of recent spawning activity). In Oklahoma, *L. blairi* enters breeding condition in spring and remains in reproductive condition into early autumn.

The range of *Lithobates blairi* (Mecham, Littlejohn, Oldham, Brown and Brown, 1973) includes parts of Indiana, Illinois, Iowa, South Dakota, Nebraska, Colorado, New Mexico, Texas, Oklahoma, Kansas, Missouri, southeastern Arizona and western Nebraska; it is a prairie grassland species (Dodd, 2013; Frost, 2019). Breeding times for *L. blairi* in the literature vary, with reproduction beginning earlier at lower latitudes and later at higher latitudes (Davis et al., 2016). The biology of *L. blairi* (as *Rana blairi*) is summarized in Brown (1992). In this paper I add to information on reproduction of *L. blairi* in Oklahoma from a histological examination of gonadal tissues. The use of museum collections for obtaining reproductive data avoids euthanizing specimens and obviates the need for a collecting permit from state and federal authorities.

A sample of 50 *L. blairi* from Oklahoma collected 1948 to 2017 (Appendix) consisting of 17 adult males (mean snout–vent length, SVL = 66.6 mm ± 10.4 SD, range = 53–85 mm); 32 adult females (mean SVL = 80.9 mm ± 13.6 SD, range = 57–100 mm) and one subadult female (SVL = 51 mm) was examined from the herpetology collection of the Sam Noble Museum, University of Oklahoma (OMNH), Norman, Oklahoma, USA. An unpaired *t*-test was used to test for differences between adult male and female SVLs (Instat, vers. 3.0b, Graphpad Software, San Diego, CA, USA).

A small incision was made in the lower part of the abdomen and the left testis was removed from males and a piece of the left ovary from females. Gonads were embedded in paraffin, sections were cut at 5 µm and stained with Harris hematoxylin followed by eosin counterstain (Presnell and Schreibman, 1997). Histology slides were deposited at OMNH.

The testicular morphology of *L. blairi* is similar to that of other anurans as detailed in Ogielska and Bartmańska (2009a). Within the seminiferous tubules, spermatogenesis occurs in cysts which are closed until the late spermatid stage is reached; cysts then open and differentiating sperm reach the lumina of the seminiferous tubules (Ogielska and Bartmańska, 2009a). A ring of germinal cysts is located on the inner periphery of each seminiferous tubule. All 17 *L. blairi* males in my sample were undergoing spermiogenesis. By month these were: March (N = 2), April (N = 4), May (N = 1), June (N = 3), July (N = 2), August (N = 1), September (N = 1), November (N = 3). The smallest mature male (OMNH 31560), measured 53 mm SVL, and was from November.

The mean SVL of *L. blairi* females was significantly larger than that of males ($t = 3.8$, df = 47, $P = 0$ .0004). The ovaries of *L. blairi* are typical of other anurans in being paired organs lying on the ventral sides of the kidneys. In adults the ovaries are filled with diplotene oocytes in various stages of development (Ogielska and Bartmańska, 2009b). Mature oocytes are filled with yolk droplets; the surrounding layer of follicular cells is thinly stretched. Monthly stages in the spawning cycle of *L. blairi* are in Table 1. Two stages were present (1) "Ready to spawn" in which mature oocytes predominated; (2) "Not in spawning condition" in which previtellogenic oocytes predominated. There were six females in the "not in spawning condition" in Table 1. One of them, OMNH 47233 (April) contained previtellogenic oocytes and partial yolk-filled oocytes. The yolking oocytes in OMNH 47233 were similar to Secondary Growth Stage 5 "progressive accumulation of yolk platelets" as reported by Uribe Aranzábal (2011). It is not known when this female would have reached spawning condition. The high percentage of *L. blairi* females in spawning condition (26/32, 81%; Table 1) reflects reproductive activity both early and late in the year. The smallest mature female (OMNH 43841) measured 57 mm SVL, was from September, and contained mature yolk-filled oocytes. One smaller female, SVL = 51 mm (OMNH 33024) contained only previtellogenic oocytes and was considered to be a subadult.

Atresia (spontaneous oocyte degeneration) is a widespread

**Table 1.** Two monthly stages in the spawning cycle of 32 adult female *Lithobates blairi* from Oklahoma.

| Month | N | Ready to spawn | Not in spawning condition |
|---|---|---|---|
| March | 7 | 7 | 0 |
| April | 7 | 6 | 1 |
| May | 7 | 6 | 1 |
| June | 3 | 1 | 2 |
| July | 2 | 1 | 1 |
| August | 2 | 1 | 1 |
| September | 3 | 3 | 0 |
| November | 1 | 1 | 0 |

**Table 2.** Months of breeding by location for *L. blairi*; * = reported as *Rana berlandieri*.

| Locality | Breeding period | Source |
|---|---|---|
| Arizona | March–June; August-October | Frost and Platz, 1983 |
| Arizona | April–June; August-October | Brennan and Holycross, 2009 |
| Colorado | Late March to August | Hammerson, 1999 |
| Illinois | March–April | Phillips et al., 1999 |
| Iowa | late March into May | LeClere, 2013 |
| Missouri | April to June, autumn? | Johnson, 2000 |
| Kansas | March to August, autumn? | Collins et al., 2010 |
| Nebraska | spring–summer | Ballinger et al., 2010 |
| Nebraska | mid–spring | Fogell, 2010 |
| New Mexico | February to October | Degenhardt et al., 2006 |
| Oklahoma* | February to September | Bragg, 1950 |
| Oklahoma | Spring–summer; autumn? | Sievert and Sievert, 2011 |
| South Dakota | March to June | Kiesow, 2006 |
| Texas | February to October | Tipton et al., 2012 |

process occurring in the ovaries of all vertebrates (Uribe Aranzábal, 2009). It is common in the amphibian ovary (Saidapur, 1978) and is the spontaneous digestion of a diplotene oocyte by its own hypertrophied and phagocytic granulosa cells which invade the follicle and eventually degenerate after accumulating dark pigment (Ogielska and Bartmańska, 2009b). See Saidapur and Nadkarni (1973) and Ogielska et al. (2010) for a detailed description of stages of atresia in the frog ovary. Atretic follicles were observed in 22% (7/32) of my mature female sample. Atresia plays an important role in fecundity by influencing numbers of ovulated oocytes (Uribe Aranzábal, 2011).

Postovulatory follicles (evidence of a recent spawning) were noted in *L. blairi* (OMNH 27545) from June. Postovulatory follicles form when the ruptured follicle collapses after ovulation; the follicular lumen disappears and proliferating granulosa cells are surrounded by a fibrous capsule (Redshaw, 1972). Postovulatory follicles are short-lived in most anuran species and are resorbed after a few weeks (Redshaw, 1972). Also present in OMNH 27545 were partially yolked follicles *sensu* Uribe Aranzábal (2011). It is not known if this *L. blairi* would have spawned again during the current year.

In conclusion, *L. blairi* reproduces primarily in spring into summer throughout its range (Table 2). The months of the spawning cycle are influenced by prevailing climatic conditions (Davis et al., 2016). Under suitable conditions, *L. blairi* may exhibit a period of autumn reproduction in some states including Oklahoma (Bragg and Dowell, 1954; Table 2). My data supports *L. blairi* reproducing in Oklahoma during autumn as evidenced by males undergoing spermiogenesis and females in spawning condition from September and November. If rainfall is heavy at any time from spring to fall, it is likely that some *L. blairi* will breed (Dodd, 2013).

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## Appendix

Fifty *L. blairi* from Oklahoma (by county) examined from the herpetology collection of the Sam Noble Museum, University of Oklahoma, Norman, Oklahoma, USA.

**Beckham:** OMNH 35584; **Blaine:** OMNH 38614; **Cimarron:** OMNH 25417, 25419, 25420, 26307, 27545, 31559, 31560–31562; **Comanche:** OMNH 33021, 33022, 33024–33026, 47207, 47213, 47215, 47216, 47220, 47221, 47225, 47227, 47229; **Cotton:** OMNH 27474; **Custer:** OMNH 42035; **Ellis:** OMNH 41714, 47232–47235, 47238; **Garvin:** OMNH 38621; **Green:** OMNH 27564, 39518; **Harper:** OMNH 25901; **Harrison:** OMNH 30137; **Jefferson:** OMNH 26269; **Kiowa:** OMNH 40355, 40359– 40361, 47239; **Marshall:** OMNH 43840, 43841; **Murray:** OMNH 31489; **Woodward:** OMNH 47240, 47241; **Woods:** OMNH 38610.

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# Kim, the Serendipitous Tiger Rattlesnake

**Roger A. Repp**
**9044 N. Valgrind Lane**
**Tucson, AZ 85743**
**repproger22@gmail.com**

It has been jokingly said that when the great creator made the Tiger Rattlesnake (*Crotalus tigris*) he saved them until last, and used the leftover parts of all the other rattlesnake species to put them together. Indeed, the tiny head, thick midriff, and huge rattle seem to defy any logical explanations of form versus function in the rattlesnake world. The why of the tiny head has been explained as an adaptation that allows the snake to fish lizards out of boulder cracks. That notion probably comes from the fact that they *do* eat lizards. But if that is the sole purpose of this dinky head of theirs, what would make *more* sense is a smaller body and rattle behind that tiny head. This would permit that same snake to crawl deeper into a crevice to get any escaping lizard. If an escaping envenomated lizard were to squeeze any further than three inches deep into a crevice, a Tiger Rattlesnake could not get to it. This tiny head being for fetching lizards from a crevice is a *ridiculous* notion. I like the spare parts idea better. But the author really can't explain the why of many things in the rattlesnake world. What I *can* do is show the reader an image of a Tiger Rattlesnake, and let you marvel with the rest of us over the why of the external features (Figure 1).

**Figure 1**. The small head and comparatively large midsection and rattle are distinguishing features of the Tiger Rattlesnake (*Crotalus tigris*). All images by the author.

Since the reader has been kind enough to look at the image in Figure 1, let's keep rocking with that particular tiger. (We will call them "tiger" from time to time in this narrative.) She was found by Blake Thomason, a rattlesnake aficionado who hails from California. Blake at times assisted Dr. Gordon Schuett and me with our rattlesnake radio-telemetry project in the Suizo Mountains. This Suizo Mountain Study has been mentioned a prodigious number of times in these columns. Should this be the first time that you have heard of the study—welcome to the CHS! The date Blake found this tiger was 18 March 2006. Gordon, Blake and I were working the far northeast side of the epicenter of our study plot, which is a smallish, stand-alone outlier hill that we call Iron Mine Hill. Through a rather comical series of coincidences, Blake found the Figure 1 tiger two meters downslope from a well known Western Diamond-backed Rattlesnake (*Crotalus atrox*) den. We were roughly 20 meters away from the den when I told Blake: "Hey Blake! There's a tiger up there by that rock pile. Go find it!" Blake dutifully went to that rock pile, and ten seconds later was hollering with *great* excitement: "Amigos! I got a tiger!" Yes sirs and madams—no lies were told. Blake found a tiger! Quoting my notes from this day, the Figure 1 tiger was first "viewed wedged in a crevice between two less than 30 cm rocks on lower SE apron edge of AD5" (AD5 is an abbreviation for Atrox Den Number 5). Blake fished this tiger out of that crevice with his salad tongs (which despite being the butt of many jokes, were highly effective as snake grabbers), and plopped her on some open ground. I snapped some images of her there, little realizing that one of these images would one day be my all time favorite tiger image. Let's move on with my notes from this day: "Captured for photos, deemed pregnant, going back to California with Blake."

Damn right amigos! Are you paying attention? On 18 March 2006, this wild tiger was "deemed pregnant" by three *Crotalus* experts. "Deemed pregnant" is really short and sweet. Those would be the two words that were inscribed in pen and ink on page 20 of my herp journal at the time. There was a *reason* to be succinct on any hallowed page of my herp journals, writer's cramp being the foremost. But you poor, *poor* reader, there is no such thing as short and sweet in a computer-assisted Roger Repp column. No way! We *must* drag this out a bit. We'll start by mentioning that the Suizo project was mostly funded from out of our own pockets. In short, we was "po' folk." If it took a dollar to go around the world, we would not have had enough money to get out of sight! Hence, we did not have the benefit of ultrasound, or any other form of high tech devices to help us with some of the trickier aspects of our study. We relied on wits, passion, knowledge and credit cards just to barely survive from year to year. But when I say this tiger was deemed pregnant in my notes, it means exactly that. We had a pregnant tiger in March. This leads to a question: When the hell did that snake *become* pregnant? I don't know. You don't either—*whoever* you are or think you may be. Let's digress some more by discussing the "deemed pregnant" aspect of the other two species of sympatric rattlesnake that we studied during the 15 year time period. We have already mentioned that Western Diamond-backed Rattlesnakes (*Crotalus atrox*) were part of the mix. The other species was the Black-tailed Rattlesnake (*Crotalus molossus*). With both of those species, we never knew that the females were pregnant until mid-June at earliest. *Never* did we look at either species in March and say "you are pregnant." Females that outwardly show their pregnancy in June makes sense if one functions under the standard paradigm of rattlesnake reproduction. A female rattlesnake found *anywhere* in the southwestern continental United States that outwardly shows being pregnant in March does *not* fit the standard picture of rattlesnake reproduction. June, yes; March, no! Before moving

**Figure 2.** (Left) The first Tiger Rattlesnake to be found on the Suizo Mountain Study Plot, 13 May 2001. She was processed, received a microchip (PIT tag), and was released soon after. (Right) The same tiger was recaptured on 22 March 2009, nearly eight years later. Her hefty distal flanks led the author to believe that she was pregnant. Where did he get that idea?

on to the next paragraph, we close the books on our Figure 1 tiger. She did indeed go back to California with Blake. She was in very good hands. She gave birth to four male offspring on 12 July 2006. We tell you this *only* to emphasize that when we deem a rattlesnake pregnant, it is.

When the good Dr. Schuett and I began the Suizo Mountain Study, we did not even know for sure that Tiger Rattlesnakes existed on our plot. I had found the shed skin of a tiger in 1992, but that wasn't the same as seeing an actual living individual. And shed skins are a rather flimsy way to prove the presence of snakes on any given patch of ground. Gordon and I began our radio-telemetry study 10 March of 2001, and on 13 May 2001—just two months later—I found that first Suizo Mountain Tiger Rattlesnake. *Crotalus tigris* # 1, or Ct1 for short, was an adult female. She was found crossing the dirt road that leads to our parking spot at 2002 hours (Figure 2, left-hand image). Her snout–vent length was 66 cm (25.98 inches), her tail length was 55 mm (2.17 inches), and her mass was 300 grams. Her rattle count was basal plus 5 segments, and segment number 6 was broken. The shape of the rattle was without taper, indicating that she was definitely a mature animal. While one can't ascertain the exact age of a rattlesnake by counting the rattle segments, the shape of the rattle reveals much about age to the experienced eye. Knowing what I know about tigers, I would guess that there is no way that she could have been less than seven years old in 2001. When I found this snake, I really, *really* wanted to put a transmitter in her. She had a lot going for her in terms of being a large and hefty prime of life female of the species. But Gordon didn't want to add a tiger to our telemetered snakes. What interested him the most with our study were the *atrox*, and in his mind, we had already diluted our *atrox* study with two Gila Monsters (*Heloderma suspectum*), and two Black-tailed Rattlesnakes (*Crotalus molossus*). In all fairness to him, at that time, he was the one providing the funding, all the tracking and lab equipment, as well as performing all the surgeries. I accepted his "no means no" answer to adding this tiger to our project, but with maximum whimpering. But the fact that he helped me to process Ct1, and even sunk one of his microchips in her (six bucks!), speaks volumes for his willingness to bend at least a little off course for the study. What I am most thankful for was that PIT tag. Despite all the bullshit I hear and sometimes *carefully* say about monitoring herps in the wild based on patterns or color, there is nothing more solid than a unique, nine-digit number that, if installed and documented properly, lasts *forever*. Ct1 was released back into the wild on 17 May 2001, and was soon forgotten. And from the remainder of 2001 until fall of 2006, our study focused on *atrox*, and a smattering of Gila Monsters.

By late summer of 2006, a quiet mutiny began to develop between Gordon and his crew. Blake Thomason began to hint that he could get us some money if I were interested in tracking tigers. I also still had one remaining credit card that was not yet maxed out. I was soon personally ordering smaller, tiger-sized transmitters. When Gordon objected, we pushed back. We wanted tigers dammit! For a rather lengthy time period, Gordon was on the verge of quitting the project. That would not have been good for our side, but we would have rocked alone regardless, because ***we wanted tigers—dammit***! After a couple years of subtle threats to take our toys and go home from all directions, Gordon reluctantly came around. By the year 2010, he even began to feign interest in what our tigers were doing. As is often the case, we were bickering over next to nothing. Yeah, we wanted to track tigers. But finding them anywhere can be a challenge, all the more so on our little hill. And the extremely fallible transmitters that we were using had a way of culling tigers as fast as we could find them. By the time 2009 rolled around, we were only tracking two of them—a male and a female. It was not until the third week in March of that year that we increased our N of transmittered tigers by 33.33%.

On the afternoon of 22 March 2009, I headed off for Iron Mine Hill. Upon arriving there, I gathered my tracking gear and headed up the west flank of the hill. First on my docket for the tracking was our male tiger #3. Midway into the experience, I had to take a time out in order to tap a kidney. While engaged in that pleasant activity, I let my eyes wander a bit. About ten meters away from me, upslope and at eye level, was a hefty

boulder that loomed above a soil pocket. It was exactly the type of situation that might harbor a Sonoran Desert Tortoise (*Gopherus morafkai*). I began to focus on that, and noted something inside that was so subtle that I had no business seeing it. At first I was telling myself: "Nah! It *couldn't* be!" But the more I stared at it, the more certain I became. I was seeing the disjointed traces of a dull orange-colored serpentine pattern. Scattered sunlight, diffused by the branches of a nearby hackberry bush, dappled the depths of the hole. Gusty winds kept the branches of that hackberry moving, which in turn set sun and shadow in a constant state of confusing motion around and upon the object in question. The turf in front of this burrow was infested with snarls of drying vines, some with faded violet-colored flowers clinging to them. The tangled vines and moving shadows further obscured the straight on view of what eventually registered in my brain as a Tiger Rattlesnake. It's a guy thing for sure, but stories of making the best finds while urinating are commonly told. Another guy thing is the inherent trait of bragging about spotting a snake in its most cryptic state. Where this tiger was concerned, if not the very best spotting that I have ever done, it was in the top five. As stated earlier, I had no business whatsoever in seeing it. Stopping for a whiz when and where I did was pure serendipity. Once the tiger was bagged, I waved my PIT tag reader over the outside of said bag. As soon as the reader beeped up a number, I went for the paper copy of our almighty "who-is-who" Suizo spreadsheet. This spreadsheet was always carried in the field with me, affixed to the same clipboard as our datasheets. (We *really* had our act together with this study). The very fact that there was a PIT tag inside this snake narrowed all possibilities, and I already knew that I had the tiger that Gordon rejected. As soon as I matched the microchip reading to the number on the spreadsheet, I began to write the following site description (exact quote from my datasheet): "034-124-556, Ct1 female first found on 5/13/01, very first tiger ever on I.M. Hill. Today, found in diffused sunlight, due west-facing tortoise hole, under 200mm thick x 1m wide x 1.2m long gneiss. Hackberry and Limber Bush near burrow, above end of Park Place Road. Mass in bag = 542g, bag = 92g, mass = 450g." Further data indicate that she was found at 1450 hours. The shaded ambient was 29°C (84°F), the hottest place close to her was 38°C (100°F). Her health received a "4"—which was the highest ranking possible for our study. Of course, the day was made with the recapture of this tiger. But I later went on to score what I call a "desert triple play" during the three short hours I spent on the hill that afternoon. A desert triple play is always a joyous occasion. That occurs when one finds a tiger, a tortoise, and a Gila Monster all in one outing. Each of these is always significant in and of itself, but like the triple play in baseball, finding all three in one trip out is a rare event.

As soon as I got this tiger home, I placed her into a previously cleaned and sanitized plastic storage box. As I gazed lovingly down on her, she was "deemed pregnant." (Figure 2, right-hand image). But there was no need for a rash judgment here, as processing this snake would reveal everything we needed to know. I arranged to have the renowned local herp vet Dr. Jim Jarchow do the transmitter implant surgery. The surgery occurred on 25 March. As soon as the snake was anesthetized, I performed the metrics on her. (Conking a snake out prior to measuring is by *far* the most accurate way to process a snake). *My* how she had grown during her eight-year absence! Her snout vent length was now 718 mm (26.27 inches), revealing growth of 58 mm, (2.28 inches). Her tail had grown by a mere 3 mm, but her mass had increased by a whopping 150 grams. Her rattle count was basal plus 7, with segment numbers 8 and 9 broken. This meant that she had shed her skin a minimum of three times during her hiatus, but the number of shedding events was probably more than that. Rattle segments commonly break off through the years, making such things as rattle counts unreliable for determining the number of ecdysis cycles over a long span of time.

Getting back to the surgery, a woman by the name of Kim Baker assisted Dr. Jarchow with the procedure. As a gesture of goodwill, I named Ct1 "Kim" in her honor, and we will stick to that name for the remainder of this article. While Kim the tiger was on the operating table, Jim gave our girl a couple gentle distal squeezes. He locked eyes with me, and announced that our newest Tiger Rattlesnake was pregnant! Hence, she had both deemed pregnant *and* proven pregnant. Hot-*diggity*-damn! As suggested earlier, Kim was not to be the first tiger who we radio-tracked, but she was to be the first *pregnant* tiger that we ever followed. She was so special to me in this regard that I gave her three days to recover from the surgery. (The norm is 24 hours. Some say it is best to get them back out sooner rather than later—within 24 hours of surgery). I released Kim on 28 March 2009. Since everything went well afterward, whatever I did, right *or* wrong in practice, yielded good results.

My first tracking session with Kim occurred on 4 April 2009 at 1235 hours. She was viewed sprawled out lengthwise as shown *in situ* in Figure 3. She was in direct sun, and her body temperature of 28.5°C (83°F) nearly matched the 29°C hot spot temperature reading. With this visit, as well as several others during the comparatively cooler weather of April, she always had her hefty hindquarters in the sun. Warming her babies? Normally, a pregnant female lying straight out in such fashion hides her delicate condition. But Kim was so distally packed that there was no way to hide it, even if she were trying. While seven days had elapsed since her release, she was found only 1.5

**Figure 3**. The tiger shown in Figure 2 received the name "Kim" while on the operating table for her transmitter implant surgery. In this *in situ* image, we see Kim as she appeared during her first tracking session, 4 April 2009.

**Figure 4**. Kim's parturition (birthing) site.

meters away from her capture spot. What I was not experienced enough with pregnant tigers to know is that they don't move much. To go into a blow-by-blow description of everything that Kim did between 4 April and 5 July 2009 would be a ponderous process. In all, 13 tracking events ensued during that 13-week time period. (We tracked everything once a week—like the weekend warriors that we were). There would have been many more hits during that time period had I known then what I know now. As briefly suggested earlier, any time that we got a visual on any of our subjects, a health assessment of 1 (thin) to 4 (fat) was always recorded on our data sheets. Kim was no different in this regard. As May rolled into June, Kim's movements could be contained within a 10-meter-diameter circle. The patch of ground this circle contained was by no means favorable to observe what was developing with her. There was a 3-meter-tall, crevice-infested, vertical mini-cliff involved to the east, and a jumble of large individual boulders to the west. To further obliterate our viewing pleasure, a dense hackberry bush and sprawling wolfberry blocked any easy access—physical *or* visual—to the vicinity (Figure 4). Hence, the visuals that we received of Kim were miserably bloody, sweat-soaked, and hurried affairs. However dreadful, we were able to get enough visuals to track her health through June. Everything we saw until 5 July indicated our highest health assessment. On 5 July came the crash in that department. Kim went from a "4" to a "1" on this night. She was viewed coiled in the entrance of a triangular-shaped opening among the west-side boulder jumble described earlier. *This* was the night that I should have gone all out to see what was going on behind and all around her. Even though Kim had dropped (I should stop there) every hint in the world that she had just given birth, I could have *puked* my disappointment when I noted the change in her body mass. The disenchantment that I felt should have been a moment of great joy and enthusiasm. But I had no clue at that moment what her mass drop implied. That was because my head was into the reproductive aspects of the Western Diamond-backed Rattlesnakes (*Crotalus atrox*) under watch. With them, we would be well-informed if we even *thought* they were pregnant by 5 July. It was not until six days later that I knew that Kim had given birth at some point prior to that 5 July visual. Figuratively speaking, Kim had caught me with my pants down with her parturition event.

On 11 July, another Suizo stalwart, Dr. Hans-Werner Herrmann, and I radio-tracked Kim and found her coiled tightly beneath a ground-hugging prickly pear cactus branch. She was now completely off Iron Mine Hill, and had moved 137 meters southeast to get there. We did something that we rarely did with our subjects, and moved her out of cover for a health assessment. As soon as we observed her emaciated rear flanks, I immediately knew that Kim had undergone parturition. A quick image taken of her distal flank has been placed alongside the image taken on 4 April to show the tremendous difference between her pre- and post-parturition condition (Figure 5). Once Kim was gently guided back into place, we hotfooted our way back to that *miserable* hackberry hell hole where she had spent all of June. Upon our arrival there, we visually scoured every crack and cranny in hopes of finding any sign of neonates. Just as we were about to call it quits, we noticed a tiny shed skin sprawled among the basal stalks of the hackberry. It was the shed skin of a neonate tiger! At the time, I was bitterly disappointed with this puny result. However, with the benefit of hindsight for all future nesting observations, Kim's birthing event was the second best in terms of giving us *any* data at all. The only wild tiger parturition event to top this one gave us a visual on one live neonate just outside the nest hole. Before our study concluded, we brought all pregnant tigers home to observe

**Figure 5**. (Left): Detail of Kim's "delicate condition" on 4 April 2009; (Right): post-parturition on 11 July 2009.

birthing. Doing that was simply the only way to get anything solid in terms of exact dates that birthing occurred, litter size, and metrics with both mom and the neonates.

As the reader can see, the image of Kim taken on 11 July 2009 (Figure 5, right side) reveals a pathetic sight. I was of course greatly troubled for her. There was little need for that concern. By 17 July, it was noted that the situation had greatly improved, and by 25 July, her health was assessed as a 3—the second highest ranking that we could give her. She entered her hibernaculum in good health on 24 October. She did not emerge from that hibernaculum until 10 April of 2010, and was looking good—but not "deemed pregnant"—when she did. By early July, she had moved completely off the hill, and established herself on the *bajada* to the south of the same. The turf she occupied is a series of undulating hillsides bisected by arroyos and min-washes. The vegetation that flanks these drainages is lush, and has small boulder fields scattered about. On 4 September 2010, Gordon Schuett, Hans-Werner Herrmann and I were entertaining four guests who were students of Dr. Rulon Clark. None of these students had ever seen a Tiger Rattlesnake in the wild. And it was not looking good for them seeing one on this day. Kim was the last tiger that we tracked, and we pinpointed her location as being under a "50mm thick by 600mm wide by 800mm long (2 inches thick, ~24 inches wide, ~31 inches long) granitic rock." This flat rock was at the edge of a sandy mini-wash, near a patch of limber bush and under the canopy of four meter tall palo verde and mesquite trees. Once again, a moment of extreme serendipity transpired, as we decided to do something that we *never* do. Since we felt that Dr. Clark's students were an extension of him, as a tip of the hat in his direction we turned that rock over so that they could see their first tiger and—***surprise***! Our eyes went out on stems when we saw not one, but two Tiger Rattlesnakes under this flat boulder (see cover image and Figure 6). The pair was not mating, but tail alignment revealed that they were very close to do doing so. The male was certainly courting Kim, and when courtship is involved with any species of snake, mating *can* follow. After we all had the chance to observe and photograph the pair, we gently lowered the boulder back into place. Whether the pair mated after that, I can't say for certain. But this event marked the very first time we had ever seen two tigers together on our study plot. In the years that followed 2009, we began to make multiple pairing/courtship observations with *Crotalus tigris*, one of which revealed full-blown coitus. All of these pairings occurred in either very late August or, as the courtship event with Kim revealed, early September.

**Figure 6**. A different perspective of Kim's courtship on 4 September 2010 (see also the cover of this issue). The male is the tiger on top.

The author wishes to take a moment to discuss the reproductive niche partitioning among the species of venomous reptiles that occupy Iron Mine Hill. We first need to be aware that when discussing the hill itself, we are describing an area that is roughly 60 meters tall by 300 meters wide (east-west) by 500 meters long (north-south). Four species of venomous reptile live on this hill. It is difficult to imagine four species of venomous reptile found *anywhere* in the continental United States within such a confined area as this. How do they pull it off? While we will never know or understand all the variables that make their successful coexistence possible, their staggered birthing processes are probably a factor. The pregnant female Tiger Rattlesnakes (*Crotalus tigris*) give birth on the hill from very late June until mid-July. Gravid Gila Monsters (*Heloderma suspectum*) lay their eggs during that same time period, but move off the hill to do so. And their young stay underground until the following May! Black-tailed Rattlesnakes (*Crotalus molossus*) give birth on the hill, but do not do so until very late July through August. And finally, the Western Diamond-backed Rattlesnakes (*Crotalus atrox*) move well into the *bajada* to give birth in August or September. If they gathered together to discuss how to best divide and conquer their turf, where parturition is concerned, they could not make a better arrangement. But the biggest mystery with the three species of rattlesnake under discussion is that all three mate during the same time period. Hence, the front end of the reproductive process seems to start at the same time, year in and year out. But the tiger somehow accelerates the reproductive process by pushing the babies out one to two months sooner than the others. It has become clear to me that nobody quite understands how this is happening. Whatever *is* happening with tigers, it all seems to work in perfect harmony with everything else.

Kim was the first Tiger Rattlesnake to show me glimmers of everything in the reproductive sense with tigers. She would have likely taught us much more were it not for a transmitter that failed prematurely. She entered her hibernaculum on 30 October of 2009. The site she entered this day had been previously used by three different *C. atrox*. In later years, several more different *atrox,* a female *C. molossus*, and a male tiger also used this site as a hibernaculum. She may have been in there with them, but due to circumstances beyond our control, we will never know. She entered this site in good health, and had been observed two months earlier with a healthy young male, as depicted on the cover of this issue. She had a strong possibility of entering this super site well on her way to being pregnant. We will never know any of these things, because on 13 February 2011, her signal died. The single most aggravating aspect of radio-telemetry is having a transmitter die prematurely, and forever losing a snake as a result. (And it certainly doesn't do the snake any good to go through life carrying nine grams of electronic junk in her innards either.)

But there is no need to end this column on a bummer note.

We have found three other tigers carrying old transmitters, and they seem to be doing fine. It wouldn't be too much of a stretch to think that Kim may still be alive. I hope so. She was a good teacher. I learned a lot from my association with her. That learning did not stop when her transmitter winked out. In fact, I think I can safely say that putting this column together brought on a new thirst for knowledge within me. I have been in deep discussions with some of our most notable authorities on rattlesnake reproduction. I am finally wrapping my brain around such terms as spermiogenesis, follicles, ovaries, oviducts, vitellogenesis and ovulation, to name a few. I should have known this stuff 20 years ago, but it's never too late to learn. I hope to one day lay out a testable hypothesis to determine when exactly tigers *do* ovulate and become pregnant. But I first must go through the raw data that exist on nearly 14 years of studying the 26 different tigers under our watch. There will be many memories, and some excellent photography to see and share along the way. If nothing else, Kim has provided the inspiration that will finally get me to mine the data on those tigers that were under our watch. Everything happens for a reason. The way that Kim came into my life, and kept revealing the subtle facts of tiger reproduction, has me believing that there *will* be more to come from her. Somehow, in some way, I expect a curtain call from Kim, our serendipitous tiger.

This here is Roger Repp, signing off from stinking hot Southern Arizona, where the turtles are strong, the snakes are handsome, and the lizards are all above average.

*Bulletin of the Chicago Herpetological Society 55(8):171, 2020*

# Herpetology 2020

In this column the editorial staff presents short abstracts of herpetological articles we have found of interest. This is not an attempt to summarize all of the research papers being published; it is an attempt to increase the reader's awareness of what herpetologists have been doing and publishing. The editor assumes full responsibility for any errors or misleading statements.

## DUSKY RATTLESNAKES IN CENTRAL MEXICO

A. Sunny et al. [2019, Herpetozoa 32:139-148] report that the dusky rattlesnake, *Crotalus triseriatus*, used to be very abundant in many parts of the highlands of central Mexico, but with the increasing human population and associated activities, the rattlesnake's habitats and populations have suffered drastic reductions and fragmentation. At the moment, the most important habitat features associated with the presence of *C. triseriatus*, the current potential distribution, and the landscape connectivity among the populations of the state of Mexico and Mexico City are unknown. Therefore, the authors used the maximum entropy modeling software (MAXENT) to analyze the current potential distribution and most important habitat features associated with the presence of the species. The variables with the highest contribution to the model were: proportion of *Abies* forest, minimum temperature of coldest month, maximum temperature of the warmest month, proportion of *Pinus* forest and annual precipitation. Furthermore, connectivity corridors were found only within mountain chains. These results highlight the necessity for conserving the patches of *Abies* forest and preserving the populations of *C. triseriatus* and the connectivity of the landscape.

## DIETS OF RUFOUS FROGS FROM CONTRASTING ENVIRONMENTS

D. J. Santana et al. [2019, Herpetozoa 32:1-6] note that despite the various impacts on animal life, some species can persist in cities by adjusting their natural histories. The rufous frog, *Leptodactylus fuscus*, is a South American species that can commonly be found in urban environments. The authors compared the diets of *L. fuscus* between an urban and a wild environment. They collected 57 individuals of *L. fuscus* and analyzed their diets, which differed significantly between the two sites. In the urban environment, Coleoptera were the prevalent prey items, whereas specimens from the wild site had a more diverse diet.

## NESTING BY BLANDING'S TURTLES

E. M. Buckardt et al. [2020, Chelonian Conservation and Biology 19(1):67-71] compiled observations of 116 nesting events by 37 female Blanding's turtles (*Emydoidea blandingii*) between 2013 and 2017 for a population located along the western shore of Lake Michigan, USA, to identify triggers of nesting activity. Across years, nesting dates were negatively related to average daily temperature during April and May, such that each degree increase in average temperature during April–May was associated with a 7-d-long shift earlier in nesting (defined as the date by which 90% of females had nested). Within years, nesting was more likely to occur when mean daily temperatures were > 18.9 °C, the moon was in its brightest phase, and wind emanated from the east or south. These thresholds may be useful for timing interventions to protect nests and nesting female Blanding's turtles, although they may differ among populations across the species' range.

## ONLINE TRADE IN CITES-LISTED TURTLES

R. W. Y. Wong et al. [2020, Chelonian Conservation and Biology 19(1):95-100] report that the Internet is being exploited as a medium for illegal wildlife trade, and protected wildlife can now be sold and bought across social media and e-commerce platforms. This article is a 13-mo study on the online trade on a localized Hong Kong website of chelonians listed under the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES). During the period of the study, more than 400 posts were collected, with more than 300 posts selling CITES-listed chelonians. Based on these findings, the authors give two general recommendations in enforcing the online illegal wildlife trade: 1) increase knowledge of CITES regulations on pet trade forums, and 2) introduce digital solutions to monitor pet trade forums.

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For sale: *Australian Reptiles & Frogs* by Raymond Hoser; 1989; 238 pp., 613 color photos; natural history, husbandry, photography, field observations, conservation, range maps; (h)—$35. *The Taipan* by Paul Masci and Philip Kendall; 1995; 89 pp., many color and b&w photos; biology, behavior, history, snakebite cases; (s)—$40. *Amphibians & Reptiles of New Mexico* by Degenhardt, Painter, and Price; 431 pp., 123 color photos (h) and *Reptiles of North Carolina* by Palmer and Braswell; 1995; 412 pp.; 76 color photos, 127 figs. (b&w drawings); (h)—both in as new condition, $37 each. *Encyclopedia of Australian Animals—Reptiles* by Harald Ehmann; 1992; 495 pp.; covers all described species with a color photo of nearly all; natural history, habitat, range maps; (h)—$75. *Snakes of Western Australia* by Storr, Smith, and Johnstone; 1986; 187 pp., 24 plates with 2–6 color photos on each; many b&w drawings; descriptions, range maps, keys; (s)—$30. (h) = hardbound, (s) = softbound. All books in excellent condition and sent postpaid. Email for complete list. William Turner, toursbyturner@aol.com.

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## NEW CHS MEMBERS THIS MONTH

Jessica Anderson
Eli D. Ehrenpreis
Kristyna Ryan
Beverly Tillman
Jordan Trevino
William R. Turner

## UPCOMING MEETINGS

**The August 26 meeting of the Chicago Herpetological Society has been canceled.**

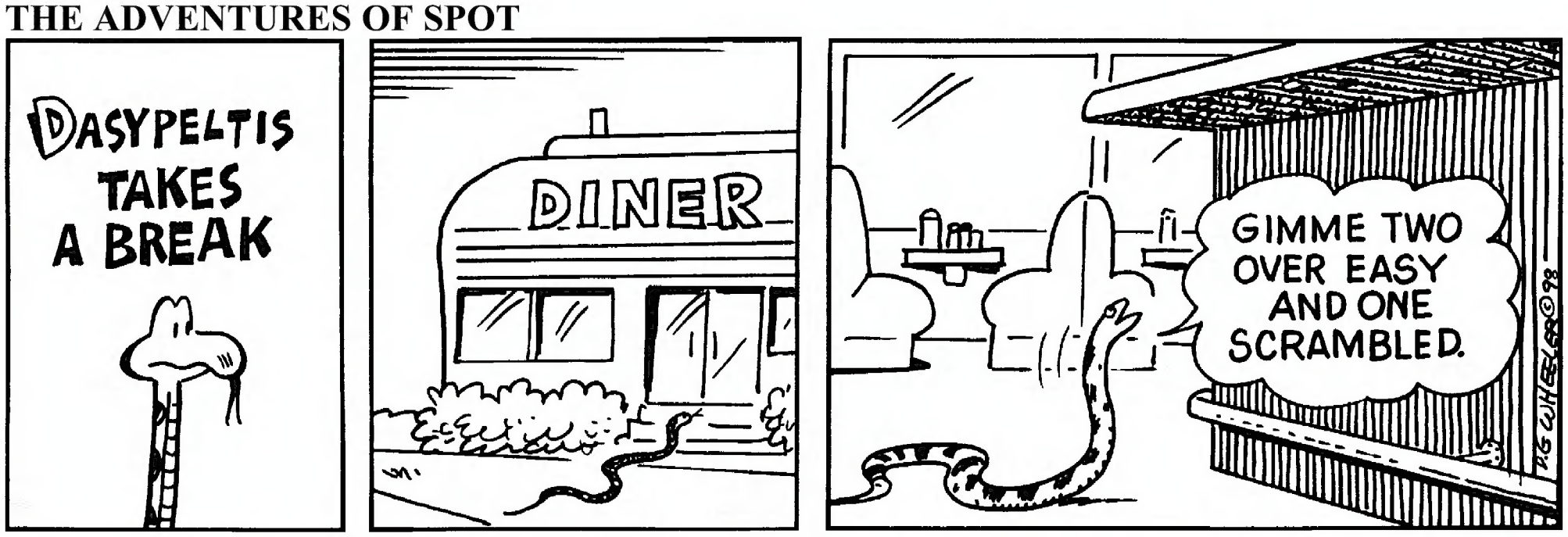
**The September 30 meeting will likely also not take place.**

The regular monthly meetings of the Chicago Herpetological Society take place at Chicago's newest museum—the **Peggy Notebaert Nature Museum**. This beautiful building is at Fullerton Parkway and Cannon Drive, directly across Fullerton from the Lincoln Park Zoo. Meetings are held the last Wednesday of each month, from 7:30 P.M. through 9:30 P.M. Parking is free on Cannon Drive. A plethora of CTA buses stop nearby.

### Board of Directors Meeting

Are you interested in how the decisions are made that determine how the Chicago Herpetological Society runs? And would you like to have input into those decisions? The next board meeting will be held online. If you wish to take part, please email mdloogatch@chicagoherp.org.

## THE ADVENTURES OF SPOT



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